

SUPPORTING INFORMATION

Supplemental Table 1: Clinical characteristics of human subjects

	Allergic	Non-allergic
Total mass cytometry samples	19	20
Male (%)	10 (53)	10 (50)
Female (%)	9 (47)	10 (50)
Total cell culture samples	14	10
Male (%)	10 (71)	6 (60)
Female (%)	4 (29)	4 (40)
Age (mean \pm SEM)	59.8 \pm 2.4	43.2 \pm 2.7
Sensitized to peanut (%)	0	0
Total IgE (kU/L) (mean \pm SEM)	361.4 \pm 128.2	66.6 \pm 17.2
Alpha-gal sIgE (kUA/L) (mean \pm SEM)	69.8 \pm 42.3	0.58 \pm 0.25

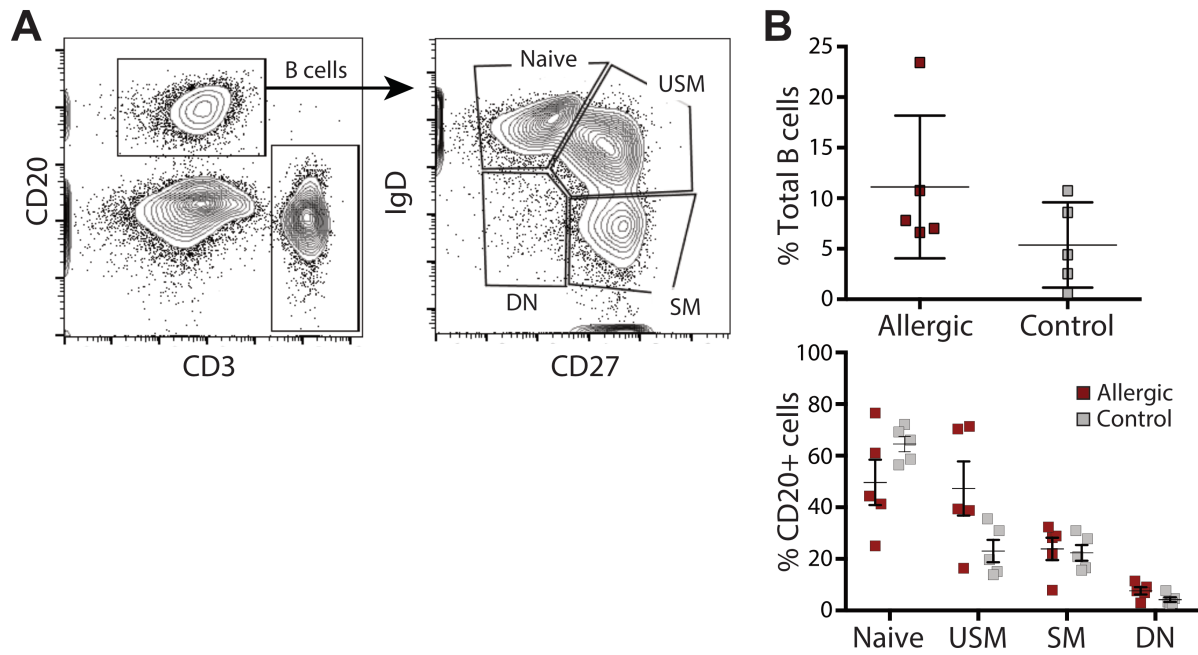
Supplemental Table 2: Mass cytometry staining panel

Antibody	Surface Molecule	Clone	Manufacturer	Label	Dilution
CD3	CD3	UCHT1	DVS Sciences	170 Er	200
CD4	CD4	SK3	DVS Sciences	174 Yb	200
CD8	CD8	SK1	DVS Sciences	168 Er	200
CD14	CD14	M5E2	DVS Sciences	151 Eu	100
CD16	FcyRIII	3G8	DVS Sciences	148 Nd	100
CD20	B lymphocyte antigen	2H7	DVS Sciences	147 Sm	200
CD24	Heat stable antigen	ML4	DVS Sciences	169 Tm	100
CD27	CD27	L128	DVS Sciences	158 Gd	100
CD38	cyclic ADP ribose hydrolase	HIT2	DVS Sciences	167 Er	100
CD43	Leukosialin	84-3C1	DVS Sciences	150 Nd	100
CD56	NCAM	CM33B	DVS Sciences	176 Yb	100
CD86	B7-2	IT2.2	DVS Sciences	156 GDd	100
CD127	IL7R- α	AO19D5	DVS Sciences	165Ho	100
CD138	Syndecan 1	DL-101	DVS Sciences	145 Nd	100
CD184	CXCR4	12G5	DVS Sciences	175 Lu	100
CD185	CXCR5	51505	DVS Sciences	171 Yb	100
CD196	CCR6	G034E3	DVS Sciences	141 Pr	100
HLA-DR	HLA-DR	L243	DVS Sciences	143 Nd	100
IgD	IgD	IA6-2	DVS Sciences	146 Nd	100
IgM	IgM	MHM-88	DVS Sciences	172 Yb	100
CD45	PTPRC	HI30	DVS Sciences	154 Sm	400
CD25	IL2R- α chain	2A3	DVS Sciences	149 Sm	100
CD11b	ITGAM	ICRF44	DVS Sciences	144Nd	100

Supplemental Table 3: Flow cytometry staining panel

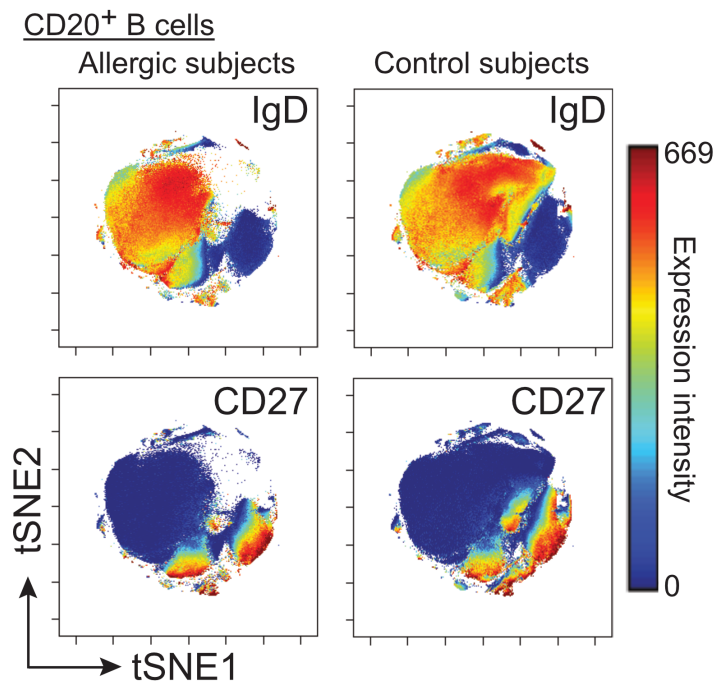
Antibody	Clone	Manufacturer	Label	Dilution
CD20	2H7	BioLegend	FITC	100
CXCR4	12G5	eBioscience	PE	50
CD43	HIT-3A	BioLegend	APC	50
CXCR5	MU5UBEE	eBioscience	PECy7	50
CD24	ML5	BioLegend	PerCPCy5.5	50
CD38	HIT2	Invitrogen	PE-Texas Red	50
IgD	IA6-2	BioLegend	APCCy7	100
IgM	SA-DA4	eBioscience	Biotin	100
HLA-DR	L243	BioLegend	BV510	20
CD25	BC96	BioLegend	BV711	20
Streptavidin	--	eBioscience	EF450	200
live/dead	--	Invitrogen	Aqua	200

Cox et al. Supplemental Figure 1



Supplemental Figure 1. Distribution of major B cell populations assessed by flow cytometry. (A and B) Mature B cells were measured by fluorescent flow cytometry. Bar plots represent mean \pm SD of the indicated cell population ($n = 5$ in each group), which were not statistically significant between groups. USM, immunoglobulin-unswitched memory B cells; SM, switched memory B cells; DN, IgD⁻ CD27⁻ double negative B cells.

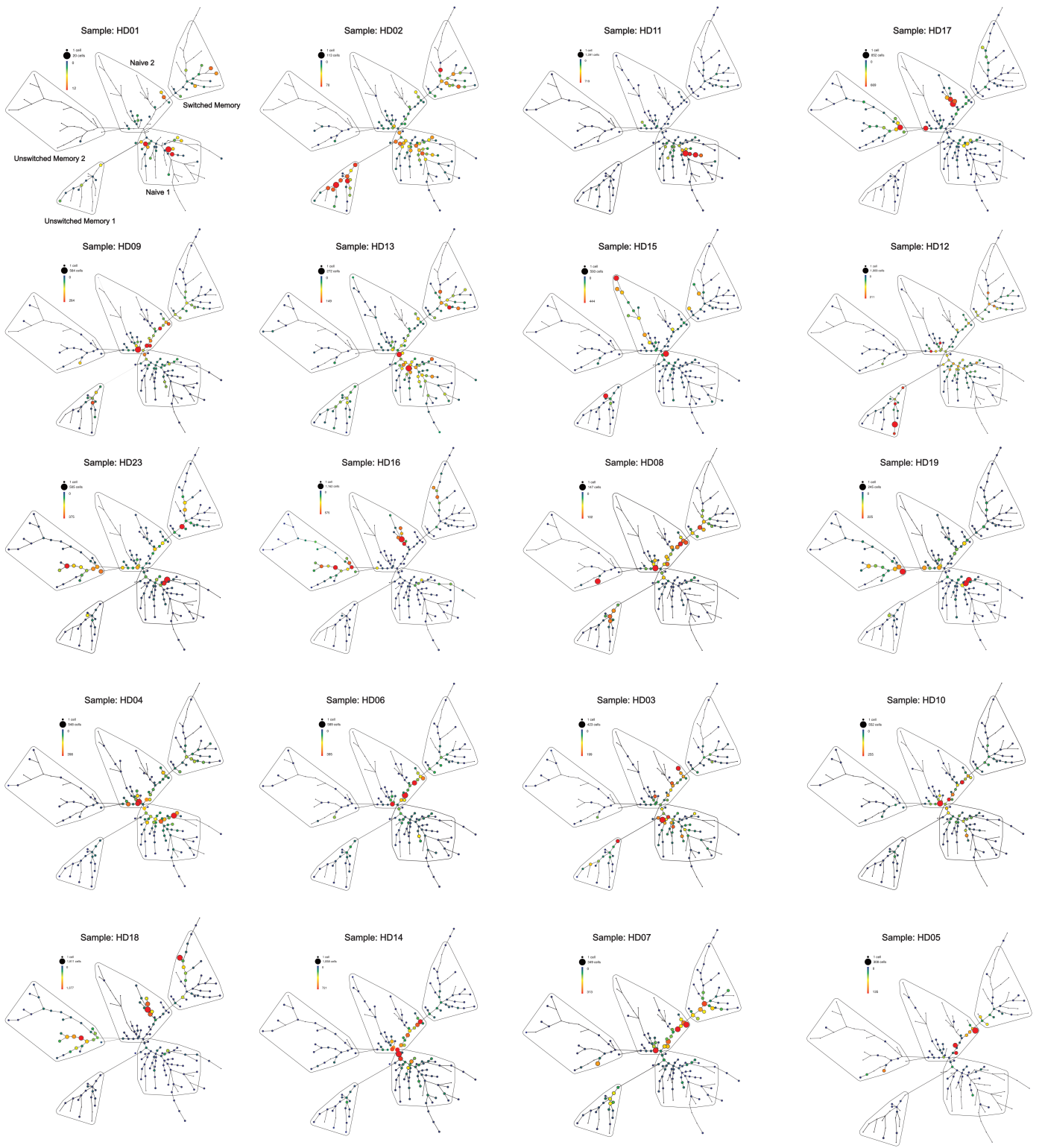
Cox et al. Supplemental Figure 2



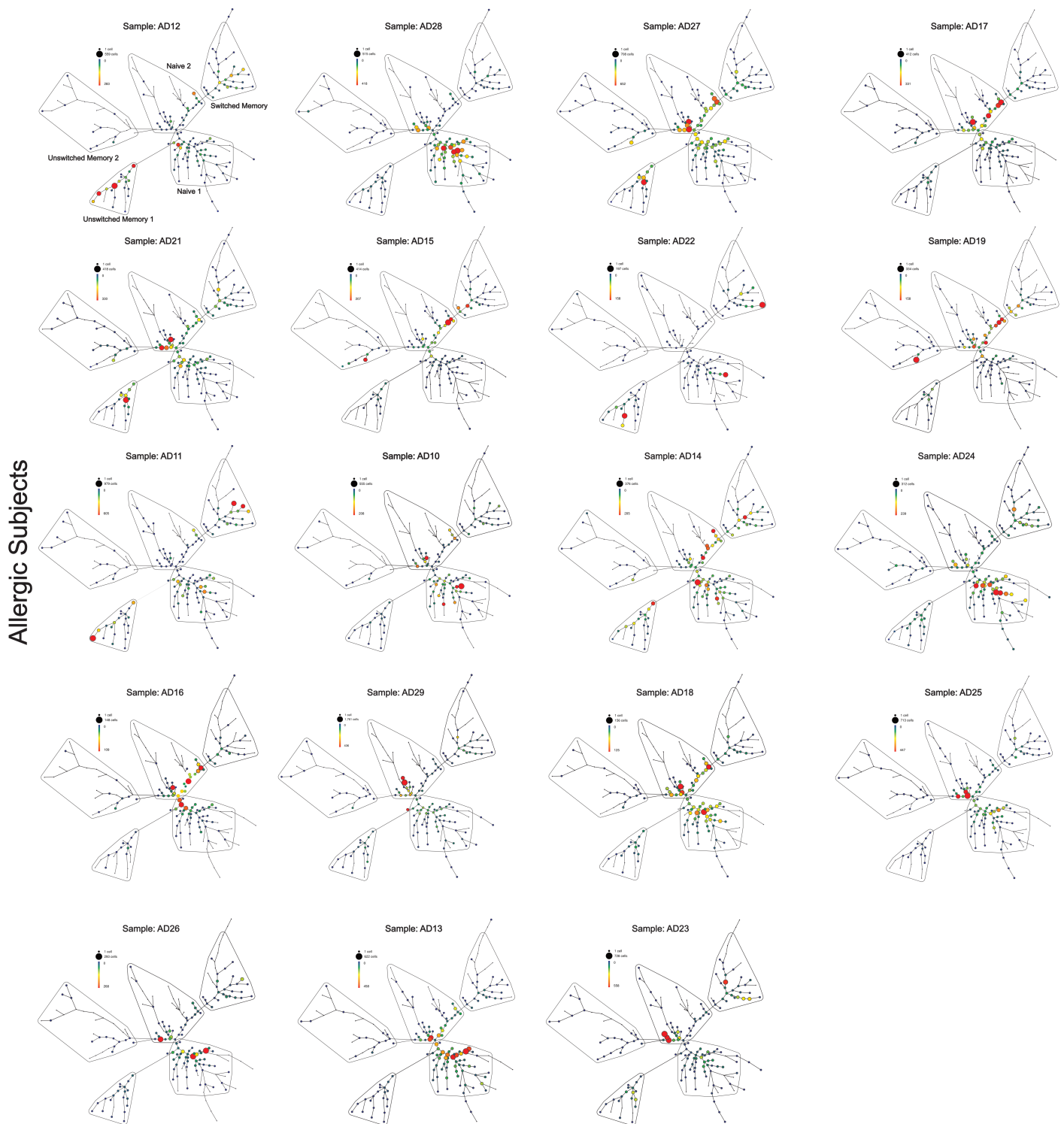
Supplemental Figure 2. viSNE maps of CD20⁺ B cells from datasets of meat allergic and control subjects. viSNE maps of B cells gated on live, singlet, CD20⁺CD3⁻ cells from concatenated datasets from meat allergic (n = 19) and non-meat allergic (n = 20) control subjects. Plots are colored by IgD and CD27 expression levels.

Cox et al. Supplemental Figure 3

Healthy Control Subjects



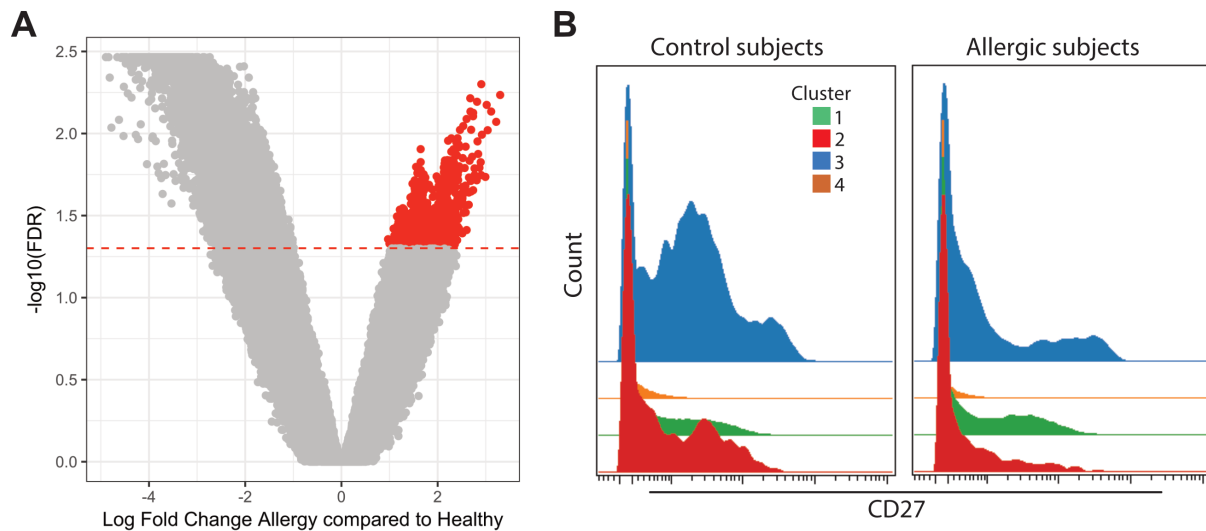
Cox et al. Supplemental Figure 3 continued



Supplemental Figure 3. SPADE analysis of CyTOF data shows phenotypic changes of B cells in meat allergic subjects. SPADE analysis of individual datasets of each subject group is shown. Each circular node represents a phenotypically similar population of B cells, with the relationship between nodes

reflecting the most similar phenotypes to adjacent nodes. Node size and color represent the number of cells. Manual gating on the basis of IgD and CD27 expression was used to annotate known B cell subsets: Naïve, (CD27⁻ IgD⁺), Unswitched memory (CD27⁺ IgD⁺) and Switched memory (CD27⁺ IgD⁻).

Cox et al. Supplemental Figure 4



Supplemental Figure 4. Assessment of changes in B cell subsets between

allergic and control subjects. (A) Volcano plot of differentially abundant B cell

immunophenotypes between 19 meat allergic subjects and 20 non-allergic controls, with negative \log_{10} -scaled FDR-corrected P-values on the Y-axis, and the \log_2 fold change on the X-axis. B cell immunophenotypes to the right and colored in red were comparatively found at higher frequencies in allergic patients. The horizontal red line indicates a significance of 0.05 after adjustment of FDR using the Benjamini-Hochberg method. Grey dots below the red line represent no changes in the frequencies of B cell immunophenotypes. B cell immunophenotypes to the left and colored in grey and above the red line were found at significantly higher frequencies in controls. (B) Histograms showing CD27 expression levels on the indicated B cell clusters measured by CyTOF.